They followed elutriated daughter cells ectopically expressing a *whi5* mutant with seven Cdk sites removed and observed a considerable extension of G1. This work was done in a different strain background, and cannot be directly compared to the experiments of Costanzo et al (2004). As a result, the detailed mechanism by which Cdk activity neutralizes the inhibitory function of Whi5 remains uncertain, but these complexities do not diminish the importance of Whi5 as a negative regulator. Rather, they remind us that cells do not rely on a single regulatory mechanism to keep their cell cycle in order. Moreover, while the general outline of cell cycle control is remarkably conserved, cells can differ with respect to which regulatory steps play the most prominent roles.

In summary, these studies extend the parallels between yeast and metazoans at the initiation of the cell cycle. Whi5 and the RB-like proteins show remarkable similarity both in their activity as repressors and in the regulation of that activity by Cdk-dependent phosphorylation. However, like the activators of transcription, there is no primary sequence homology between Whi5 and RB. Rather, these parallels serve as an exquisite example of convergent evolution and irrefutable testimony to the logic of this circuitry. They also illustrate the power of comparative studies in diverse model organisms for expediting a full understanding of any fundamentally conserved process.

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# Catching Some Zs: A New Protein for Spatial Regulation of Bacterial Cytokinesis

**Prior to its duplication, the bacterial nucleoid exerts local negative control over assembly of the cytokinetic Z ring to prevent potential cutting of the chromosome. In this issue of *Cell*, Wu and Errington show that a specific nucleoid-associated protein mediates this nucleoid occlusion effect, providing the first mechanistic insight into this key spatial regulatory system.**

The choice of where to divide is a fundamental problem for most cells and determines the fate of progeny cells. In the case of rod-shaped prokaryotes such as *Escherichia coli* or *Bacillus subtilis*, the positioning of the cytokinetic ring at the cell midpoint is negatively regulated by the centrally positioned nucleoid. This effect, called nucleoid occlusion (NO), prevents unwanted cutting of an unsegregated chromosome by the cell division septum (Woldringh et al., 1990). The primary target of NO appears to be the tubulin-like FtsZ protein, which forms the cytokinetic Z ring. In mutant cells defective for chromosome segregation or replication initiation, the Z ring localizes to the edge of the nucleoid, far from the cell center (Harry, 2001; Margolin, 2001). This remarkable mislocalization indicates that the Z ring can potentially assemble anywhere in the cell but responds to negative effectors by assembling where the negative effect is lowest.

Another negative spatial regulator of cytokinesis is also found in many bacteria. Consisting of MinC, MinD, and MinE or DivIVA proteins, the Min system prevents unwanted Z ring assembly in nucleoid-free areas at the cell poles (Migocki et al., 2002). In Min$^-$ cells, Z rings form at all DNA-free areas of the cell, including correct positions between daughter nucleoids and incorrect positions at the cell poles, but do not form over nucleoids because of NO (Yu and Margolin, 1999). Min$^-$ cultures are viable because sufficient septation occurs medially to produce many daughter cells with chromosomes. The Min system exerts its effect via MinC, which locally inhibits FtsZ polymer assembly. This inhibition is spatially controlled by the action of the MinD protein, the majority of which is mainly restricted to the polar portion of the cell membrane by MinE in *E. coli* (Raskin and de Boer, 1999) or DivIVA in *B. subtilis* (Marston et al., 1998).

The currently accepted model for Z ring placement is that prior to chromosome segregation, the independent action of NO and Min-mediated inhibition results in the masking of all points on the cell membrane for Z ring assembly. Only when NO is relieved at the cell center during chromosome segregation is the medial Z ring allowed to assemble. Unfortunately, up to now, the mechanism behind NO has remained mysterious. Local DNA concentration appears to be an important mediator because conditions that reduce chromosome condensation result in suppression of NO, with Z rings often assembling on top of nucleoids (Margolin, 2001). The stage of chromosome replication is also an important factor (Harry, 2001). However, a molecular handhold has been needed for some time to gain a better understanding of how NO works.

A breakthrough has come with the discovery by Wu and Errington (2004) of a protein that mediates NO in *B. subtilis*. They found, serendipitously, that a mutant in *yyaA*, previously shown to have no discernible phenotype, was lethal when combined with a *min* mutant. To determine the cause of this lethality, they found that when both *yyaA* (subsequently called *noc* for "nucleoid occlusion") and *min* were inactivated, cells stopped dividing. Instead of forming rings exclusively between

nucleoids, FtsZ in the double mutant often localized in multiple patches on top of normal-looking nucleoids. This suggested that NO was specifically suppressed and that Noc was important for NO.

But why was cell division inhibited in the double mutant, which removed two negative regulators? One potential model to explain this apparent paradox proposes that normal assembly of the Z ring requires molecular crowding (Rivas et al., 2001), which might tend to occur when most of the membrane surface is masked by NO and the Min system. When one system is missing, the other would still mask sufficient space. However, when both systems are absent, the limited amount of cellular FtsZ fails to assemble productively because there would be too many potential assembly sites. This effect was observed previously with an *ftsZ84 min* double mutant, in which a weakened FtsZ actually assembled less well in the absence of Min inhibition (Yu and Margolin, 2000). As with the *ftsZ84 min* mutant, overproduction of FtsZ in *noc min* double mutants resulted in partial suppression of the division inhibition phenotype, which supports the hypothesis.

If Noc is so important for nucleoid occlusion, then why did a *noc* null mutant lack detectable defects? To address this, Wu and Errington (2004) perturbed the cell cycle in order to uncover important roles for Noc in mediating NO. When cells were made longer by inhibiting later steps of cell division, Z rings, normally restricted to internucleoid spaces, assembled promiscuously on top of nucleoids near the center of these long cells. This supports the idea that Min-mediated inhibition of Z ring assembly extends a considerable distance from the cell poles but at some point is unable to block assembly on top of nucleoids in the absence of Noc. In the most dramatic demonstration of the importance of Noc, blocking reinitiation of chromosomal DNA replication caused division septa to cut nucleoids at high frequency in *noc* mutants, while they pinched to one side of nucleoids in $Noc^+$ cells.

How might Noc function to inhibit Z ring assembly? Noc is weakly homologous to the ParB/Spo0J family of proteins, and like these, Noc localizes to nucleoids. However, whereas ParB/Spo0J localize to polar foci near the partitioned chromosomal origin, Noc localizes to a more dispersed portion of the nucleoid. Interestingly, Noc did not localize to the middle of segregating nucleoids, which correspond to replication termini near the cell midpoint. A gradient of lower NO near the terminus, along with lower inhibition by the Min system at the center of a long predivisional cell, might be sufficient to permit assembly of the Z ring at the cell center at the correct time in the cell cycle. Perhaps assembly of the asymmetric Z ring on top of the filamentous nucleoid during *B. subtilis* sporulation is also mediated by a local decrease in Noc localization. Conversely, overproduction of Noc decreased the frequency of Z rings and partially inhibited cell division, possibly by localizing to lower affinity sites near the terminus or even away from the nucleoid.

The major challenges now will be to determine if Noc acts directly on FtsZ, how Noc blocks assembly of the Z ring, what other factors influence FtsZ positioning, and whether NO and Noc are widespread. Many prokaryotes lack Min homologs, and unless they have other proteins that perform a similar function, they may rely solely on NO. This idea makes sense if their nucleoids fill the cells, preventing significant DNA-free gaps at the cell poles. *Caulobacter crescentus,* for example, lacks a Min system, but Z rings tend to localize in areas of the cell with low DNA concentrations (Quardokus and Brun, 2002), suggesting that NO plays an important role. While NO may be universal, Noc is probably not, as it is poorly conserved at the sequence level. Clearly, the discovery of Noc delivers NO out of the realm of phenomenology and confirms the importance of NO. Its discovery also ushers in a new era for understanding the regulation of prokaryotic cellular organization.

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# Siah Proteins, HIF Prolyl Hydroxylases, and the Physiological Response to Hypoxia

**New evidence suggests that at least two members of the family of hypoxia-inducible factor (HIF) prolyl hydroxylases that regulate HIF stability in response to oxygen ($O_2$) availability are also targeted for proteosome-dependent degradation by the E3 ubiquitin ligases Siah1a and Siah2. This preview examines cellular responses to $O_2$ deprivation (hypoxia) and the complexity of the regulation of the HIF $O_2$ sensing pathway in mammals.**

Oxygen homeostasis is maintained in higher eukaryotes via highly developed respiratory and circulatory systems, as $O_2$ is central to the viability of most organisms. Conserved $O_2$ sensing pathways are present in all mammalian cells and tissues and critical for a variety of physiological and pathological processes (reviewed in Semenza [2000]). Many rapid intracellular responses to low $O_2$ (hypoxia) exist, but hypoxia-inducible transcription factors (HIFs) regulate a majority of the changes in gene